Claims

1. A method of analyzing cells disposed in media within a vessel, the method comprising the steps of:

a) providing a vessel having an original volume of media about the cells;

b) reducing the original volume of media about at least a portion of the cells to define a reduced volume of media; and

c) analyzing a constituent related to the cells within the reduced volume of media.

2. The method of claim 1, further comprising the step of increasing the reduced volume of media about the cells to substantially the original volume.

3. The method of claim 1, further comprising the steps of:

determining a first concentration of the constituent; and

determining a second concentration of the constituent at a predetermined time interval from determining the first concentration.

4. The method of claim 3 further comprising the step of calculating a flux rate of the constituent based on the first concentration and the second concentration.

5. The method of claim 1 wherein the reduced volume is in a range of about 5-50% of the original volume.

6. The method of claim 5 wherein the reduced volume is in a range of about 5-20% of the original volume.

7. The method of claim 1 wherein the reduced volume is less than about 5% of the original volume.

8. The method of claim 1 wherein the cells comprise a cell selected from the group consisting of bacteria, fungus, yeast, a prokaryotic cell, a eukaryotic cell, an animal cell, a human cell, and an immortal cell.

9. The method of claim 1 wherein at least a portion of the cells are attached to a surface of the vessel.

10. The method of claim 1 wherein at least a portion of the cells are suspended in the media.

11. The method of claim 1 wherein at least a portion of the cells comprise living tissue.

12. The method of claim 1 wherein the constituent comprises a material selected from the group consisting of a dissolved gas, an ion, a protein, a substrate, a salt, and a mineral.

13. The method of claim 12 wherein the dissolved gas is selected from the group consisting of O_2, CO_2, and NH_3.

14. The method of claim 12 wherein the ion is selected from the group consisting of H^+, Na^+, K^+, and Ca^{++}.

15. The method of claim 12 wherein the protein is selected from the group consisting of a cytokine, insulin, a chemokine, a hormone, and an antibody.

16. The method of claim 12 wherein the substrate comprises a material selected from the group consisting of glucose, a fatty acid, an amino acid, glutamine, glycogen, and pyruvate.

17. The method of claim 1 wherein the constituent comprises a material extracted from the media by at least a portion of the cells.

18. The method of claim 1 wherein the constituent comprises a material secreted into the media by at least a portion of the cells.

19. The method of claim 1 wherein analyzing the constituent comprises sensing presence of the constituent.

20. The method of claim 1 wherein analyzing the constituent comprises sensing concentration of the constituent.

21. The method of claim 1 wherein analyzing the constituent comprises sensing a first concentration of a first constituent, sensing a second concentration of a second constituent, and determining a relationship between the first concentration and the second concentration.

22. The method of claim 1 wherein analyzing the constituent comprises sensing a rate of change of concentration of the constituent.

23. The method of claim 1 wherein analyzing the constituent comprises using a sensor in contact with the media within the reduced volume.

24. The method of claim 23 wherein the sensor comprises a sensor selected from the group consisting of a fluorescent sensor, a luminescent sensor, an ISFET sensor, a surface plasmon resonance sensor, a sensor based on an optical diffraction principle, a sensor based on a principle of Wood's anomaly, an acoustic sensor, and a microwave sensor.

25. The method of claim 1 wherein analyzing the constituent comprises determining a parameter selected from the group consisting of cell viability, cell number, cell growth rate, response to at least one of a drug, a toxin, and a chemical, detection of an entity, and internalization.

26. The method of claim 1, further comprising the step of perfusing additional media through the vessel.

27. The method of claim 1, further comprising the step of replenishing the media in the vessel.

28. The method of claim 1 wherein reducing the volume of media comprises disposing a barrier in the vessel.

29. The method of claim 28 wherein the barrier is disposed in the vessel without causing displacement of media out of the vessel.

30. The method of claim 28 wherein at least a portion of the barrier comprises a sensor.

31. The method of claim 28 wherein the reduced volume of media comprises a sensor.

32. The method of claim 31 wherein the sensor comprises a fluorophore.

33. The method of claim 1 wherein at least a portion of the vessel comprises a sensor.

34. The method of claim 1, further comprising the step of altering an environment of at least a portion of the cells prior to reducing the original volume of media.

35. The method of claim 34 wherein altering the environment comprises exposing at least a portion of the cells to at least one of a drug, a chemical, and a toxin.

36. The method of claim 1, further comprising the step of altering an environment of at least a portion of the cells after reducing the original volume of media.

37. The method of claim 1, further comprising the step of covering the vessel.

38. The method of claim 1, further comprising the step of stirring at least a portion of the original volume of media in the vessel.

39. The method of claim 1, further comprising the step of sealing the vessel.

40. An apparatus for analyzing cells, the apparatus comprising:

a) a stage adapted to receive a vessel holding cells and a volume of media;

b) a plunger adapted to receive a barrier to create a reduced volume of media within the vessel including at least a portion of the cells, the barrier adapted for insertion into the vessel by relative movement of the stage and the plunger; and

c) a sensor in sensing communication with the reduced volume of media, wherein the sensor is configured to analyze a constituent disposed within the reduced volume.

41. The apparatus of claim 40 wherein the sensor is configured to analyze the constituent without disturbing the cells.

42. The apparatus of claim 40 wherein the vessel comprises a well disposed in a microplate.

43. The apparatus of claim 42 wherein the well comprises a step.

44. The apparatus of claim 40 wherein the barrier is adapted to stir the media prior to analysis of the constituent.

45. The apparatus of claim 40 wherein the sensor is selected from the group consisting of a fluorescent sensor, a luminescent sensor, an ISFET sensor, a surface plasmon resonance sensor, a

sensor based on an optical diffraction principle, a sensor based on a principle of Wood's anomaly, an acoustic sensor, and a microwave sensor.

46. The apparatus of claim 40 wherein at least a portion of the vessel comprises the sensor.

47. The apparatus of claim 40 wherein the reduced volume of media comprises the sensor.

48. The apparatus of claim 40 wherein at least a portion of the barrier comprises the sensor.

49. The apparatus of claim 40, further comprising an automated electro-optical measurement system.

50. The apparatus of claim 49, further comprising a computer in electrical communication with the automated electro-optical measurement system.

51. The apparatus of claim 40 wherein the barrier is biased relative to the plunger.

52. The apparatus of claim 40 wherein:

the vessel comprises a microplate having a plurality of wells; and

the barrier comprises a plurality of barriers arranged to be received within a plurality of the wells.

53. The apparatus of claim 52, wherein the plurality of barriers are independently biased relative to the plunger.

54. An apparatus for analyzing cells, the apparatus comprising:

a) a stage adapted to receive a vessel holding cells and a volume of media;

b) a plunger adapted to receive a barrier to create a reduced volume of media within the vessel including at least a portion of the cells, the barrier adapted for insertion into the vessel by relative movement of the stage and the plunger without disturbing the cells, such that the reduced volume is less than about 50% of the volume of media;

c) a sensor in sensing communication with the reduced volume of media, wherein the sensor is configured to analyze a constituent disposed within the reduced volume.

55. A plate comprising multiple wells adapted for holding media and cells, each of at least a portion of the wells comprising a seating surface adapted for receiving a barrier configured to define a reduced volume.

56. The plate of claim 55 wherein a shape of the seating surface is selected from the group consisting of generally planar, arcuate, contoured, tapered, conical, stepped, and interlocking.

57. The plate of claim 55 wherein the reduced volume within each of the wells varies by less than about 10% of a mean volume of the wells.

58. The plate of claim 57 wherein the reduced volume within each of the wells varies by less than about 5% of the mean volume of the wells.

59. The plate of claim 58 wherein the reduced volume within each of the wells varies by less than about 1% of the mean volume of the wells.

60. The plate of claim 55 wherein the seating surfaces of the wells each comprise a step disposed about an inner periphery of a respective well.

61. The plate of claim 60 wherein the steps lie in a step plane disposed above a bottom plane defined by bottoms of respective wells.

62. The plate of claim 60 wherein the step plane and the bottom plane are parallel planes.

63. The plate of claim 62 wherein a height of the step plane is less than about 1 mm above the bottom plane.

64. The plate of claim 63 wherein the height of the step plane is less than about 200 μm above the bottom plane.

65. The plate of claim 64 wherein the height of the step plane is less than about 50 μm above the bottom plane.

66. The plate of claim 55, further comprising a fluorescent sensor disposed within at least one of the wells.

67. The plate of claim 55 wherein at least one of the wells comprises a transparent bottom.

68. The plate of claim 55 wherein at least one of the wells comprises an opaque wall.

69. A barrier for analysis of cells disposed in media in a vessel, the barrier comprising:
a body portion for insertion into the vessel, the body portion having a barrier surface for mating with a first surface of the vessel to create a reduced volume.

70. The barrier of claim 69 wherein the barrier surface comprises a shape selected from the group consisting of generally planar, arcuate, contoured, tapered, conical, stepped, and interlocking.

71. The barrier of claim 69, further comprising a cover for mating with a second surface of the vessel.

72. The barrier of claim 69, further comprising a sensor disposed on the barrier surface for analyzing a constituent of a media disposed about at least a portion of the cells.

73. The barrier of claim 72 wherein the sensor comprises an optical sensor.

74. The barrier of claim 73 wherein the optical sensor is adapted to sense a fluorophore.

75. The barrier of claim 73, further comprising a conductor coupled to the sensor adapted to conduct signals therefrom.

76. The barrier of claim 75 wherein the conductor comprises an optical fiber.

77. The barrier of claim 75 wherein the conductor is disposed at least partially in the body portion.

78. The barrier of claim 72, further comprising a readout for transmitting a signal from the sensor.

79. The barrier of claim 78 wherein the readout is selected from the group consisting of visual, fiber, electronics on a post, and a plate reader from the bottom.

80. The barrier of claim 69, wherein the barrier comprises a plurality of barriers arranged to be received within a plurality of wells in a microplate.